SPECTRAL PROPERTIES OF SOME WEST COAST WATERS AND THEIR RELATIONSHIP WITH DISSOLVED ORGANIC CARBON.

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ABSTRACT

Water samples were collected from 40 West Coast stream and river sites and analysed for dissolved organic carbon (DOC) and absorbance at 320nm. Extinction coefficients (E_{430}) and E_4/E_6 ratios were calculated for samples from 22 of these sites. DOC concentration ranged from 1.6-43.2g m^{-3} and was related linearly to absorbance at 320nm. The equation describing the relationship (DOC (g m^{-3}) = $34.98A_{1cm}$ + 1.55) was comparable to equations found by other workers at similar wavelengths. A curvilinear relationship was found between DOC concentration and E_4/E_6 ratios (range 1.8-14.0) suggesting that proportionately more low molecular weight humic substances were present in brown waters than in clear waters. Extinction coefficients ranged from 0.0005 to 0.0047 and were correlated significantly with E_4/E_6 ratios indicating that low molecular weight humic substances absorbed more light per unit organic carbon than humic materials of higher apparent molecular weight.

KEY WORDS: Dissolved organic carbon, absorbance, extinction coefficient, E_4/E_6 ratio, aquatic humic substances

INTRODUCTION

In most aquatic ecosystems dissolved organic carbon (DOC) is the dominant form of organic carbon in the water column. A major fraction of DOC (50-75%) is made up of humic substances which generally are characterised as yellow-coloured, organic compounds (humic, fulvic and hydrophilic acids) that are refractory end products in the degradation of plant and microbial organic matter (Thurman, 1985). Natural DOC concentrations in streams and rivers normally range from 1-10g m^{-3}. However, concentrations in wetlands can be much higher (average 30g m^{-3}) because of the large net primary production of emergent plants and the presence of slow moving streams that leach vegetation and interstitial soil water (Thurman, 1985).

The colour of humic substances is caused by the presence of multiple bonds and unshared electron pairs in the molecular structure (Antherton et al., 1967; Orlov, 1972). Water colour can be measured spectrophotometrically and many workers have used absorbance of water at a specific wavelength as an index of DOC concentration (e.g., Banoub, 1973; Lewis and Canfield, 1977; Grieve, 1985). Absorption characteristics of humic substances also can be expressed by extinction coefficients and colour quotients (sensu Visser, 1984) such as E_4/E_6 ratios. Extinction coefficients give a measure of the degree of absorptivity per unit organic carbon and may be used to compare samples from different localities (Thurman, 1985). Colour quotients are related to the steepness of the slope in a spectrogram between two predetermined wavelengths and are usually independent of DOC concentration. E_4/E_6 ratios are believed to reflect particle size of the humic material and thus are indirect indicators of molecular weight (Chen et al., 1977).

In a recent paper (Collier, 1987) I assessed the utility of absorbance at 360nm as a measure of DOC concentration in 40 streams and rivers on the west coast of South Island, New Zealand. The present study examines other spectral properties (extinction coefficients and E_4/E_6 ratios) of water samples from some of these sites and evaluates the use of absorbance as an index of DOC concentration in the light of recent findings by other workers.

METHODS

SAMPLE COLLECTION:

Water samples (250mL, n=47) were collected from 40 stream and river sites on the west coast of South Island between February and May 1986. Sites 2,3, and 4 (Appendix 1) were the only streams sampled more than once. A brief description of the study area and map references of the sampling sites are given in Collier (1987). Samples were kept cool in the dark in polyethylene bottles until their return to the laboratory where they were filtered (0.45µm) and analysed, normally within one week of collection.

DOC ANALYSES:

Subsamples of water (50 or 100 mL) were evaporated to dryness and DOC concentration was determined by the micro dichromate oxidation procedure (Maciolek, 1962; 0.05N $K_2Cr_2O_7$, 0.03N $FeSO_4.7H_2O$) using heat by dilution (the heat generated on mixing sulphuric acid and the dichromate solution). DOC concentration was calculated according to the method of Maciolek (1962):

$$(1)\quad \text{DOC (mg)} = \frac{(A-B) \times N \times 8}{2.86}$$

where A= mL of ferrous sulphate used in blank titration
B= mL of ferrous sulphate used in sample titration
N= normality of ferrous sulphate
8= equivalent weight for oxygen
2.86= factor converting the weight of oxygen to organic carbon

Normality of ferrous sulphate was calculated by titration against the potassium dichromate solution:

$$(2)\quad N = \frac{\text{mL dichromate} \times \text{normality of dichromate}}{\text{mL ferrous sulphate used in titration}}$$

The technique was calibrated by ashing beech (Nothofagus) detritus and tannic acid (Mallinkcrodt analytical reagent) overnight at 500°C and comparing ash free dry weight with the amount of organic carbon extracted by dichromate oxidation. Organic matter was assumed to be 50% carbon (Maciolek, 1962).

SPECTROPHOTOMETRIC ANALYSES:

Absorbance spectra (200-650nm) of 4 representative water samples (Sites 3,10,29, and 37) and absorbance at 320nm (all samples) were measured using a Kontron Uvicon 860 spectrophotometer (1cm quartz cell). Absorbances of water samples from 22 of the sites were measured at 430, 460, and 660nm in a 10cm glass cell (Varian DMS 100 spectrophotometer). Distilled water blanks were used when making all absorbance measurements. E_4/E_6 ratios were obtained from the quotient of absorbance at 460 and 660nm and extinction coefficients were calculated by dividing absorbance at 430nm (per cm of cell path length) by the weight of organic carbon (g m^{-3}) in the sample. Gjessing (1974) proposed 430nm as a standard wavelength for comparing water colour. However, wavelengths used by other workers for calculating extinction coefficients vary from 400nm (Thurman, 1985) to 540nm (Visser, 1984)

RESULTS

DOC ANALYSES:

(a) Oxidation efficiency

Calibration of the heat by dilution dichromate oxidation procedure showed that it retrieved an average of 83% of the organic carbon in tannic acid and beech detritus (Table 1). Maciolek (1962) recommended immersing the acid/ dichromate mixture in a boiling water bath for 3 hours to facilitate maximum oxidation of the organic carbon. This step was omitted in the present study to enable rapid analysis of a large number of samples. DOC concentrations calculated from Equation 1 therefore needed to be multiplied by 1.20 to provide a more accurate estimate of total DOC (Table 1). The equation used to calculate DOC can be simplified to:

(3) DOC (mg) = (A-B) x N x 3.36

Lee et al. (1983) used heat by dilution dichromate oxidation to measure the organic carbon content of some South Pacific soils. They compared carbon values obtained by this method with estimates obtained using a high frequency induction furnace and found that corrections equivalent to those used in Equation 3 were necessary to give accurate estimates of organic carbon content.

(b) DOC concentrations in stream waters

DOC concentrations of water samples collected from the 40 sites ranged from 1.6 to 43.2g m^{-3} and were highest in streams draining pakahi (wetland) catchments in Larrys Creek Experimental Area (Sites 2-4), north of Reefton (Appendix 1). DOC concentrations at most sites (70%) were below 10g m^{-3} and at 48% of the sites were less than 5g m^{-3}.

SPECTROPHOTOMETRIC ANALYSES:

Absorbance (A) at 320nm was related linearly to DOC concentration (r^2=0.92). The relationship is expressed by the equation:

(4) DOC (g m^{-3}) = $34.98A_{1cm}$ + 1.55

Spectra of the four representative water samples all showed an increase in absorbance with decreasing wavelength (Fig. 1). The rate of increase was proportional to DOC concentration and was most pronounced in the ultra violet range of the spectrum (<360nm). Extinction coefficients at 430nm (E_{430}) for the 22 samples analysed ranged from 0.0005 to 0.0047 and 77% of the values were between 0.002 and 0.004 (Appendix 1). The lowest E_{430} and E_4/E_6 ratio were found in the water sample from Waiho River (Site 40) which drains Franz Josef glacier.

A curvilinear relationship was found between DOC concentrations and E_4/E_6 ratios (Fig. 2). All samples with ratios less than 7.0 had DOC concentrations below 5.2g m^{-3}. The highest ratios (13.5 and 14.0) were recorded in water samples from Sites 2 and 3 (L1 and L2, Appendix 1).

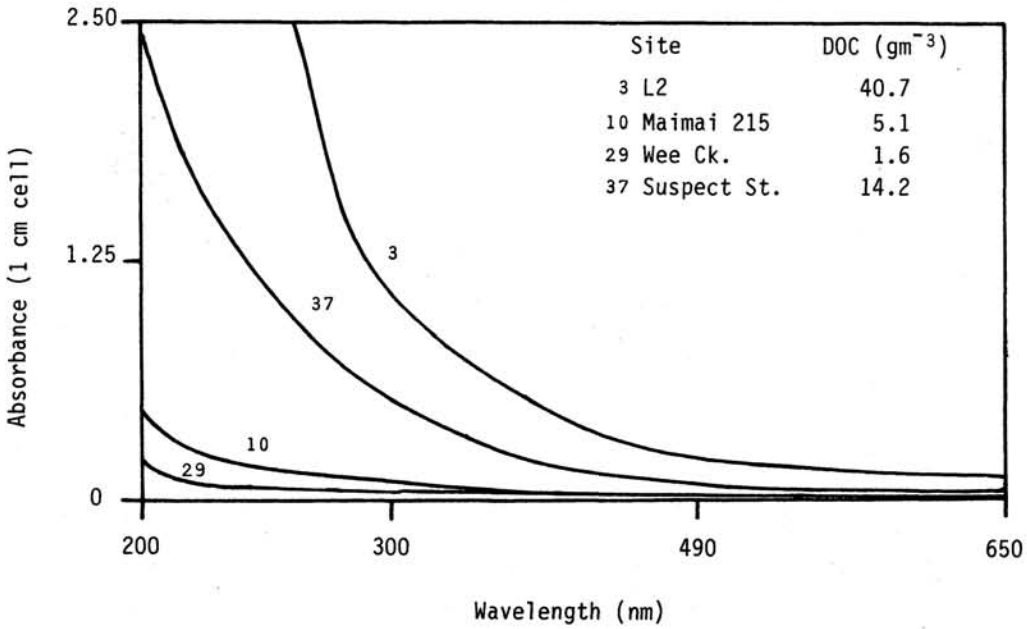

Fig. 1 Absorbance spectra of water samples from 4 sites with dissolved organic carbon (DOC) concentrations ranging from 1.6 to 40.7g m^{-3}.

DISCUSSION

Absorbance spectra for the four West Coast water samples resemble spectra obtained by Moore (1985) for peat water samples from northern Quebec, Canada. The undifferentiated nature of these spectra is the result of a multitude of overlapping excitation levels caused by very large numbers of chromophores in the humic molecule (Visser, 1984).

TABLE 1 Percent organic carbon recovered from tannic acid and beech detritus using heat by dilution dichromate oxidation. The right hand column gives factors by which organic carbon values obtained by the Maciolek (1962) calculation must be multiplied to correct for inefficiency in oxidation. AFDW = initial ash free dry weight of the compound oxidised.

Compound	AFDW (mg)	% recovery	Multiplication factor
Tannic acid	17.6	80.6	1.24
	20.0	85.1	1.18
	19.0	84.2	1.19
Beech detritus	28.7	86.2	1.16
	28.7	82.6	1.21
	28.7	79.1	1.26
± SD		83.0 ± 2.7	1.20 ± 0.03

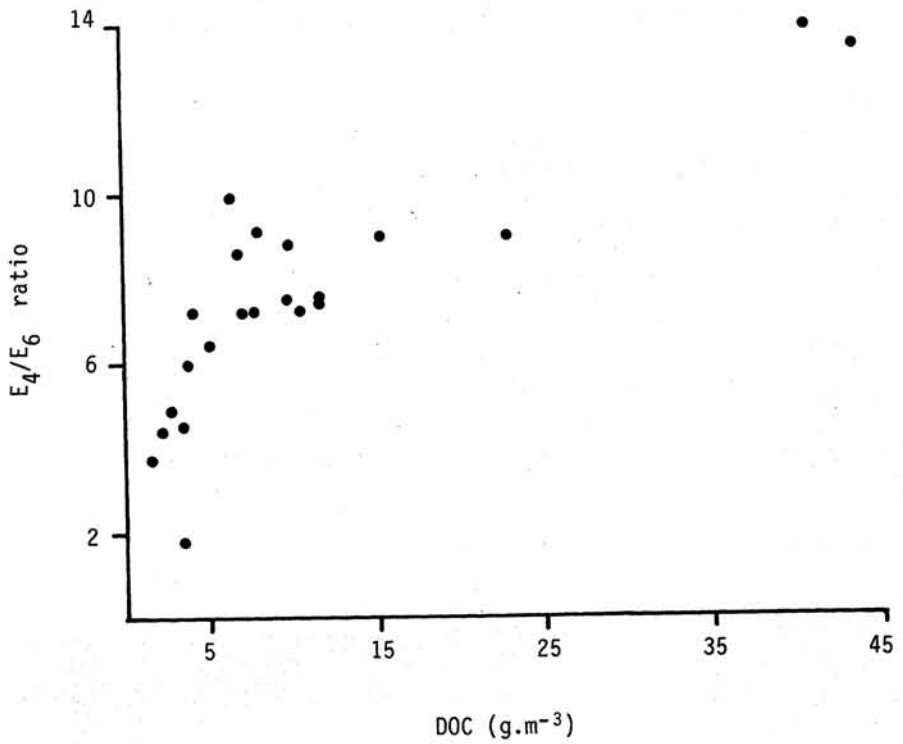

Fig. 2 Relationship between dissolved organic carbon (DOC) concentrations and E_4/E_6 ratios of water samples collected from 22 streams and rivers on the west coast of South Island.

Regression equations expressing relationships between DOC concentration and absorbance at 320 or 330nm in several studies including this one are given in Table 2. Slopes of all equations are similar (34.98-45.49) but intercepts vary from 1.39 to 7.46. The equation expressing the relationship between DOC concentration and absorbance at 360nm for Sites 1-40 is very similar to that given by Lewis & Canfield (1977) who measured absorbance at 360nm in some Venezuelan and southeastern United States waters (see Collier, 1987).

Chen et al. (1977) observed that water pH had a pronounced effect on E_4/E_6 ratios which reached maxima between pH 6 and 8. The pH of the West Coast water samples used to calculate E_4/E_6 ratios ranged from 4.1 to 7.8 (Appendix 1). In this pH range, Chen et al. (1977) showed that E_4/E_6 ratios changed by no more than 0.5 units and, thus, water pH is unlikely to have affected ratio estimates in the present study. Using a temporally extensive data set (24 weekly samples), Moore (in press) recorded mean E_4/E_6 ratios of 7.3 for Sites 5-10 and 11.1 for Sites 2-4. These compare favourably to 7.4 and 12.2, respectively, calculated from spot samples taken in the present study. Several workers have shown that E_4/E_6 ratios are related inversely to molecular weight of the humic material (Butler & Ladd 1969, Chen et al. 1977, Visser 1984). Thus, high ratios such as those recorded in samples from Sites 2 and 3 (13.5 and 14.0, respectively) infer enrichment by low molecular weight humic substances relative to samples with smaller ratios. Chen et al. (1977) recorded an E_4/E_6 ratio of 13.7 at pH 4 for a fulvic acid fraction with a molecular weight of 833, and all fractions with ratios less than 8 had molecular weights greater than 2109. Increasing DOC concentrations in the West Coast waters sampled appeared to be associated with decreases in the apparent molecular weight of the humic material.

TABLE 2 Details of dissolved organic carbon (DOC)/ absorbance relationships at 320 and 330nm from contrasting natural waters. A = absorbance in a 1cm cell.

Source	Wavelength (nm)	DOC range ($g\ m^{-3}$)	r^2	Equation (DOC ($g\ m^{-3}$)=)
This study	320	1.6-43.2	0.92	34.98A + 1.55
Gorham & Detenbeck 1986[1]	320	16.7-83.9	0.73	42.8A + 5.1
Moore 1985[2]	330	1-51	0.84	45.05A + 1.80
Moore in press[3]	330	2-18	0.92	45.49A + 1.39
Moore in press[4]	330	18-55	0.88	38.87A + 7.46

1 North American bog water
2 Canadian peat water
3 Sites 5-10 of this study
4 Sites 2-4 of this study

Stewart & Wetzel (1981) noted that dissolved organic matter (DOM) of low molecular weight absorbed consistently more light than DOM of higher molecular weight. They suggested that this was a function of molecular geometry whereby strongly absorbing hydrophilic groups of high molecular weight humic substances are restricted spatially to central portions of the molecule where their absorbing characteristics are masked partially by more external, lower-absorbing hydrophilic groups. However, Visser (1984) did not detect a significant relationship between absorbance per unit carbon (extinction coefficients) and molecular weight of aquatic humic substances from lakes, rivers, streams and swamps.

In the present study, extinction coefficients were related significantly to E_4/E_6 ratios ($r=0.56$, $P<0.01$), indicating that absorptivity was a function of the apparent molecular weight distribution of aquatic humic substances. Waters with high DOC concentrations had higher E_4/E_6 ratios and appeared to be dominated by lower molecular weight humic materials than clear waters where E_4/E_6 ratios were smaller.

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LITERATURE CITED

ANTHERTON, N.M., CRANWELL, P.A., FLOYD, A.J. & HAWORTH R.D. 1967. Humic acid. I. E.S.R. spectra of humic acids. *Tetrahedron 23*: 1653-1667.

BANOUB, M.W. 1973. Ultraviolet absorption as a measure of organic matter in natural waters in Bodensee. *Archiv fur Hydrobiologie 71:* 159-165.

BUTLER, J.H.A. & LADD, J.N. 1969. Effect of extractant and molecular size on the optical and chemical properties of soil humic acids. *Australian Journal of Soil Research 7:* 229-239.

CHEN, Y., SENESI, N. & SCHNITZER, M. 1977. Information provided on humic substances by E_4/E_6 ratios. *Soil Science Society of America Journal 41:* 352-358.

COLLIER, K.J. 1987. Spectrophotometric determination of dissolved organic carbon in some South Island streams and rivers. *New Zealand Journal of Marine and Freshwater Research 21.*

GORHAM, E. & DETENBECK, N.E. 1986. Sulphate in bog waters: a comparison of ion chromotography with Mackereth's cation- exchange technique and a revision of earlier views on cause of bog acidity. *Journal of Ecology 74:* 899-903.

GRIEVE, I.C. 1985. Determination of dissolved organic matter in streamwater using visible spectrophotometry. *Earth Surface Processes and Landforms 10:* 75-78.

GJESSING, E.T. 1974. Physical and chemical characteristics of aquatic humus. Ann Arbor, Michigan. Ann Arbor Science publ. 120p.

LEE, G.W., SINGH, U. & MORRISON R.J. 1983. The determination of organic carbon in South Pacific soils. *South Pacific Journal of Natural Science 4:* 34-42.

LEWIS, W.M. & CANFIELD, D. 1977. Dissolved organic carbon in some dark Venezuelan waters and a revised equation for spectrophotometric determination of dissolved organic carbon. *Archiv fur Hydrobiologie 79:* 441-445.

MACIOLEK, J.A. 1962. Limnological organic analysis by quantitative wet oxidation. United States Fisheries and Wildlife research report no. 60. 61p.

MOORE, T.R. 1985. The spectrophotometric determination of dissolved organic carbon in peat waters. *Soil Science Society of America Journal 49:* 1590-1592.

MOORE, T.R. in press. An assessment of a simple spectrophotometric method for the determination of dissolved organic carbon in freshwaters. *New Zealand Journal of Marine and Freshwater Research.*

ORLOV, D.S. 1972. Modern chemical and physical techniques for studying the nature and structure of soil humic substances. *Soviet Soil Science 1972:* 461-467.

STEWART, A.J. & WETZEL, R.G. 1981. Assymetrical relationships between absorbance, fluoresence and dissolved organic carbon. *Limnology and Oceanography 26:* 590-597.

THURMAN, E.M. 1985. Organic geochemistry of natural waters. Dordrecht. Martinus Njhoff/ Dr W. Junk Publishers. 497p.

VISSER, S.A. 1984. Seasonal changes in the concentration and colour of humic substances in some aquatic environments. *Freshwater Biology 14:* 79-87.

APPENDIX 1 pH, dissolved organic carbon (DOC) concentrations, extinction coefficients (E_{430}) and E_4/E_6 ratios of water samples collected from 22 West Coast sites in February 1986. Grid references of sites are given in Collier (1987). -, unnamed streams.

Site no.	Name	pH	DOC ($g\ m^{-3}$)	E_{430}	E_4/E_6
1	Buller R.	6.2	2.3	0.0026	4.4
2	L1	4.1	43.2	0.0035	13.5
3	L2	4.1	40.7	0.0042	14.0
4	L3	4.3	22.9	0.0027	9.0
5	Maimai 205	5.7	11.5	0.0033	7.4
6	Maimai 207	6.1	10.4	0.0028	7.2
7	Maimai 208	5.8	9.6	0.0024	7.5
8	Maimai 209	6.0	7.0	0.0031	7.2
9	Maimai 214	5.8	6.8	0.0018	8.6
10	Maimai 215	5.8	5.1	0.0027	6.4
11	Inangahua R.	6.6	4.1	0.0033	7.2
22	Lankey Ck.	6.8	7.8	0.0030	7.2
26	Inangahua R.	6.2	9.7	0.0031	8.8
27	Rahu R.	6.4	3.6	0.0038	4.5
28	Inangahua R.	5.5	11.6	0.0030	7.6
29	Wee Ck.	7.5	1.6	0.0031	3.7
30	Lewis R.	7.1	2.8	0.0027	4.9
31	-	4.5	15.4	0.0047	9.0
32	Clear Ck.	6.5	3.7	0.0033	6.0
33	Hercules Ck.	5.1	8.1	0.0038	9.1
39	-	4.7	6.6	0.0043	9.9
40	Waiho R.	7.8	3.5	0.0005	1.8